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A PRELIMINARY STUDY OF ERGOT OF WILD RICE

FAITH FYLES

WITH PLATE XI

In my experiments with the cultivation of Canadian wild rice, *Zizania aquatica* L. & *Z. palustris* L., I had often noticed ergot among the seed. A search of the literature revealed only one record of the occurrence of ergot on wild rice (R. H. Denniston (1)).

The questions arose, first, is this ergot identical with the typical ergot (*Spermoedia clavus*(D. C.) Fries), of rye, second, will cereals and other grasses growing in its neighborhood be in danger of infection by it?

Dr. Robert Staeger (2) of Bern, Switzerland, who has been successfully experimenting with ergot of rye for many years, has already proved that ergot, which cannot be distinguished morphologically, may, however, require quite a different range of host plants. He has shown by a series of successful experiments that ergot of rye, *Secale cereale*, will infect the following grasses:

Anthoxanthum odoratum, *Hierochloe odorata*, *Poa pratensis*, *Phalaris arundinacea*, *Arrhenatherum elatius*, *Dactylis glomerata*, *Poa sudetica*, *Festuca pratensis*, *Festuca arundinacea*, *Barley*, *Hordeum murinum*, *Alopecurus pratensis*, *Briza media*, *Poa hybrida*, *Poa compressa*, *Calamagrostis arundinacea*, *Bromus sterilis*, *Secale cereale*, Spanischer Doppelroggen.

He has further shown that *Claviceps microcephala* Tul., (now *Spermoedia microcephala* (Wallr.) Seaver) of which the typical host plant is *Phragmites communis* Trin., does not exist on a single host plant subject to attacks of *Spermoedia clavus* (D. C.) Fries. *S. microcephala* infects, he states, besides its host, *Nardus stricta*, *Molinia coerulea*, and *Aira caespitosa*.

I secured a small quantity of fresh wild rice seed, gathered at Treadwell, Ontario, September 29, 1913. Among the seed were many ergots, several of which were still within the glumes of the wild rice. On October 9, 1913, most of these sclerotia were placed in pots of sand outside the laboratory window. Occasionally they were watered until they became buried in snow. Others of the same lot of sclerotia were placed in sterilized sand and water in two stender dishes kept in the laboratory at some distance from the window. They were given water twice, once in November and once in December. On December 16, two sclerotia had already germinated. One sclerotium had 19 stromata from 1 to 5 mm. high and the other showed 11 stromata. Judging from later observations, germination must have taken place a week or ten days before, i.e., a little over two months after the sclerotia were gathered. Although these ergots had been kept at room temperature from September 29, they had already, in the open air, been subjected to 2° frost on September 16, 1913.

The ostiola of the perithecia appeared in the form of minute dots on December 22 when the stromata were only 7 mm. high and the heads 1 mm. in diameter. They gradually became more distinct as the perithecia protruded until finally the stromata looked like diminutive spiked clubs. The first spores were seen on December 30. There were 40 stromata at one time on this sclerotium, the most mature being 2.5 cm. high with a head 2.5 mm. wide, while the youngest could just be seen through a crack in the cortex. One other ergot of this sowing produced 40 stromata and a third 37. The remainder did not germinate so freely, the last showing only 10 heads. Of later sowings, the greatest number of heads on a single intact ergot was 48, and the least 11. Broken pieces of ergots produced from 3 to 7 stromata. The heads were light buff in color with lavender stalks, becoming deeper and slightly reddish with age. At the height of maturity, the perithecial chambers measured 250 to 325 μ in length and 150 to 160 μ in width. The asci were 200 to 215 μ long and about 4 μ wide. The spores were 150 to 180 μ long.

On reading H. T. Güssow's account (3) of tri-septate spores recorded in his ergot of barley, I examined the spores of my fungus and am able to confirm his finding of three septa in each spore, which evidently indicates a generic character.

As wild rice is essentially an aquatic plant and naturally most of the ergots would drop into the water to float away or be washed ashore, more sowings were made: In unsterilized sand and water; unsterilized mud and water and in distilled water alone. In the first experiment, the sclerotia germinated abundantly and produced strong and healthy heads. Those in mud and water with about 2 mm. of water above the mud flourished also, and grew 4.5 to 5 cm. high with heads 2.5 mm. wide. Those floating on the distilled water germinated freely on all sides of the sclerotia, but their growth was slow, and, when they were only 5 mm. high, they became covered with mould fungi. The mould was removed as far as possible, and the sclerotia were transferred to sterilized sand and water. A few of these stromata finally produced spores, but most of them perished before maturity.

In all these sowings, most of the ergots were placed merely on top of the sand or mud. A few were buried an inch or more, but not one of the latter germinated. Even those which were brought to the surface after being buried three months did not show signs of life.

On May 22, 1914, the last of the number of sclerotia, which had been left exposed all winter outside were divided; one-half was brought into the laboratory and the other half left in a box of mud and water beside the growing wild rice. On June 30, when those in the laboratory had almost finished their spore-shedding and were 4 to 4.5 cm. high, those outside had just begun to shed their spores. This was before the rice had shown any signs of blooming. The growth was very thick and short—only 1 cm. high with heads 3 to 3.5 mm. in diameter and of a dark reddish purple color, indicating the strong positive heliotropism to which Claviceps is known to be subject.

The following table gives the data of the different stages of growth taken from one or two sclerotia of each sowing; all harvested in September, 1913.

INFECTION EXPERIMENTS WITH ERGOT OF WILD RICE

At the time of the first sowing of ergot of wild rice, October 9, 1913, we had just received a small barrel of wet wild rice seed of the autumn's gathering. In this were found a dozen seeds or more that had already germinated. As this autumnal germination of wild rice is very unusual, the young seedlings were carefully planted in large glass jars with 15 cm. of mud. The jars were then filled with water and placed, some in the laboratory and some in the greenhouse for infection experiments. Later, twelve pots of rye and barley were sown for the same purpose. But the wild rice grew slowly in the winter and the barley and rye did not make the desired progress. It was necessary to make repeated sowings of ergot all through the winter in order to have on hand mature stromata when the grasses bloomed. When, at last, the first stalks of rye and barley were in bloom, the spore-shedding of one entire lot of ergot was over and the next lot was not yet mature.

Unfortunately, at that time, I had not read Dr. Staeger's (4) recent work on infection experiments with ergot of rye, in which he showed that the conidia still adhering to the wintered sclerotia of ergot of rye were capable of germination and infection even after ten months' rest. On reading this paper, I at once re-examined the ergot, which had been kept dry in a stoppered glass jar, and found several ergots with a whitish coating, some of which were still within the glumes of the wild rice. The glumes were carefully removed and a dry white substance was found at the base of the ergots as well as inside the glumes. These ergots and glumes were soaked in distilled water for a few minutes. The water at once became milky; a drop under the microscope proved it to be full of conidia.

TABLE 1

Data on the development of sclerotia harvested September 1913

DATE OF SOWING	SOWN IN	FIRST APPEARANCE OF STROMATA	FIRST APPEARANCE OF PERITHECIA	HEIGHT AT THAT TIME	FIRST EMERGENCE OF SPORES	HEIGHT AT THAT TIME	NO. OF STROMATA ON THE SCLEROTIUM	GREATEST HEIGHT OF STROMATA
1913		1913		*millimeters*		*millimeters*		*millimeters*
Oct. 9	Sterilized sand and water	Dec. 10	Dec. 22	7	Dec. 30	20	40	25
Oct. 9	Sterilized sand and water	Dec. 18	Dec. 22	10	Dec. 31	29	37	30
Oct. 9	Sterilized sand and water	Dec. 18	Dec. 23	12	Jan. 2	20	40	23
		1914						
Oct. 9	Sterilized sand and water	Feb. 4	Feb. 14	15	Feb. 20	25	15	28
Oct. 9	Sterilized sand and water	Feb. 4	Feb. 14	15	Feb. 26	23	10	23
		1913						
Dec. 16	Sterilized sand and water	Dec. 23	Jan. 2	10	Jan. 7	12	27	15
Dec. 16	Sterilized sand and water	Dec. 23	Jan. 2	10	Jan. 8	12	23	20
Dec. 16	Sterilized sand and water	Dec. 29	Jan. 11	15	Jan. 15	20	25	21
		1914						
Dec. 16	Sterilized sand and water	Mar. 6	Mar. 16	8	Mar. 24	10	26	15
Dec. 16	Sterilized sand and water	Mar. 6	Mar. 18	12	Mar. 28	14	25	21
Dec. 17	Unsterilized mud and water	Jan. 5	Jan. 13	8	Jan. 19	20	36	45
Dec. 17	Unsterilized mud and water	Jan. 5	Jan. 20	13	Jan. 25	27	29	50
		1913						
Dec. 17	Unsterilized sand and water	Dec. 31	Jan. 13	5	Jan. 22	24	22	40
		1914						
Jan. 3	Sterilized sand and water	Feb. 2	Feb. 13	10	Feb. 19	15	38	20
Jan. 3	Sterilized sand and water	Feb. 4	Feb. 16	11	Feb. 26	17	15	24
Feb. 20	Sterilized sand and water	Mar. 14	Mar. 31	10	Apr. 14	17	48	20
Feb. 20	Sterilized sand and water	Mar. 17	Mar. 31	9	Apr. 13	15	34	18
Feb. 20	Sterilized sand and water	Mar. 18	Apr. 2	9	Apr. 11	10	46	15
Mar. 28	Sterilized sand and water	Apr. 14	Apr. 30	10	May 4	13	14	10
Mar. 28	Sterilized sand and water	Apr. 17	May 2	10	May 6	14	11	17
May 4	Sterilized sand and water	May 23	June 2	10	June 10	23	40	30
May 4	Sterilized sand and water	May 25	June 3	10	June 11	23	40	30
May 4	Sterilized sand and water	May 27	June 3	10	June 12	25	23	35

First inoculation of barley and rye

The barley and rye spikes, which were then at anthesis as well as those a little past this stage, and others in which the kernel had begun to form, were all inoculated with this spore-containing solution on April 24, 1914. No honeydew appeared on any of them, not even after a month. Only healthy seeds were seen on the now ripened heads.

First inoculation of wild rice

In the meantime, the wild rice in the greenhouse had bloomed. In the afternoon of May 11, fresh ascospores of the ergot were caught in water and sprinkled on the pistillate flowers of one stalk of wild rice, pollinated that morning. On May 15, the flowers were still very fresh, and, fearing they had been inoculated too soon and determined to obtain some positive result from this experiment, I re-inoculated them, both with fresh ascospores and with conidia obtained from the wintered ergots. Thus, when honeydew did appear, it was uncertain whether the conidia or ascospores were the cause of it. On the morning of May 30, 1914, two large drops of honeydew were found on this stalk of wild rice. A little of it in a drop of water was examined under the microscope, and found to be full of conidia. They measured 9 to 12 μ in length and 2.5 to 3.5 μ in width. (Those of *S. clavus* are 7 μ in length and 3.5 μ in width.)

On June 1, there were four drops of honeydew. The lowest drop, which was the first to appear, had changed somewhat in appearance, being now less clear and more viscid. On June 3, there were unmistakable signs of two purplish sclerotia. June 6, honeydew had ceased oozing, and three large and firm sclerotia were seen. June 11, as the sclerotia and also the plant seemed to be drying prematurely, the stalk bearing the ergots was cut off to preserve it. A little of the fresh honeydew had previously been secured in a small bottle.

Second inoculation of wild rice

On June 1, 1914, a second jar of wild rice in another wing of the greenhouse bloomed for the first time. This was inoculated with the spore-containing water of the dry ergots of September 1913.

On June 11, a drop of honeydew appeared on the uppermost flower and in two days the beginnings of a sclerotium were visible. On June 17, a lower flower also showed honeydew. The success of this second infection experiment with wild rice, which had only been inoculated with last year's conidia, proves that the conidia are still capable of germination even after nine months' rest, which confirms Dr. Staeger's discovery in regard to the ergot of rye.

EXPERIMENTS WITH RYE AND OTHER HOSTS OF ERGOT OF RYE

The rye plants, which were inoculated at the same time as the wild rice, i.e., May 11, 1914, with fresh ascospores and May 15, with old conidia, still showed no signs of honeydew.

On May 30, when honeydew first appeared on wild rice, a little of it was used to inoculate the last pot of rye in bloom. With some more of this same fresh honeydew, on June 6, the following grasses, which had been brought in from the experimental plots outside before they had bloomed, were inoculated. *Poa pratensis*, *Dactylis glomerata* and *Alopecurus pratensis*, which are all hosts of the ergot of rye. On June 8, rye growing out in the open, and on June 10 and 11 *Arrhenatherum elatius* and more rye in the greenhouse, also brought in from outside, were inoculated with the spore-containing solution of the sclerotia of September 1913, but without any positive result.

On June 12, thirty-five stalks of rye in bloom were inoculated with old conidia—with negative results.

July 3, three more stalks of rye then in bloom were treated with fresh conidia, but without any appearance of honeydew.

A number of oats in flower, also specimens of *Elymus dasystachys* and *Agropyron tenerum* were inoculated repeatedly with old and fresh conidia without any positive result.

It may be said that the study of this form of ergot proved most interesting. The rapidity of the development of stromata and the large number observed make this ergot a most desirable one for class purposes. From certain morphological differences, the difference in the measurements of conidia and ascospores, the fact that plants readily infected with spores of *S. clavus* (D. C.) Fries, proved immune in my experiments, and from some other biological features, I feel justified in considering this ergot a distinct species, and in proposing for it a new specific name. On completion of my work, the usual diagnosis will accompany the final account.

In conclusion, I wish to acknowledge my indebtedness to H. T. Güssow, the Dominion Botanist, for his kindness in editing the manuscript and making timely and important suggestions.

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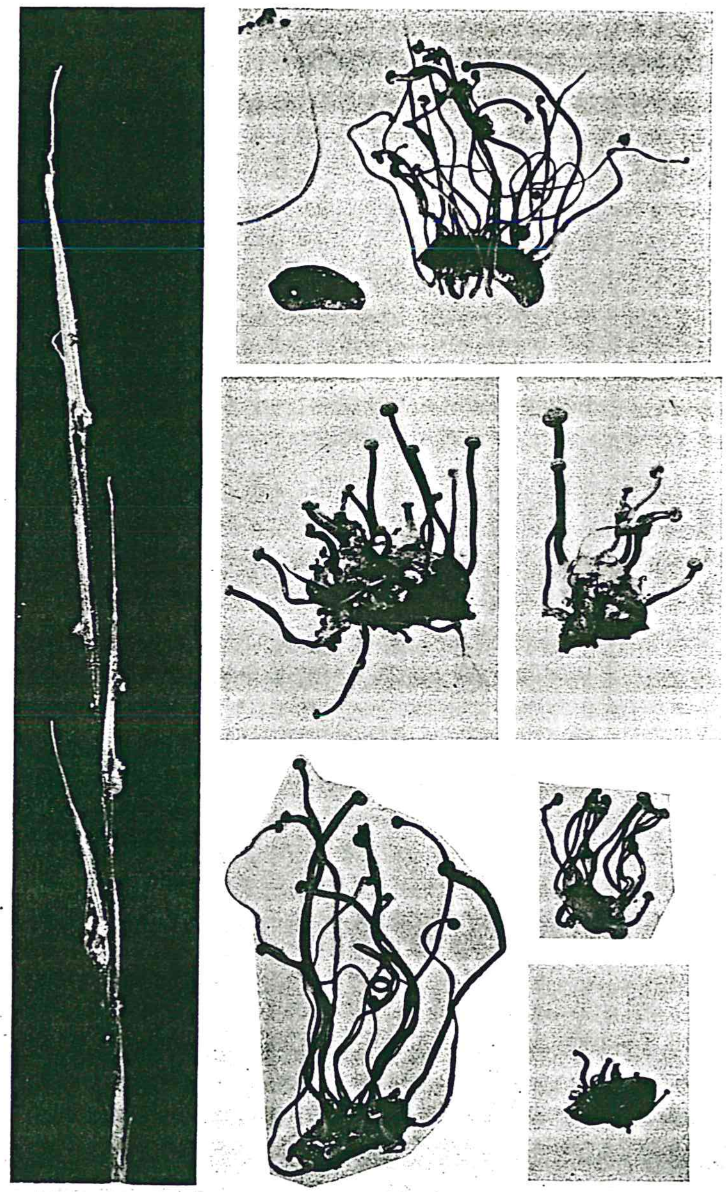
EXPLANATION OF PLATE XI

Artificially infected flowers of wild rice.
Various stages of germinating ergots.



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